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PERSISTENT TIDAL CYCLES OF SPONTANEOUS MOTOR ACTIVITY IN THE FIDDLER CRAB, *UCA PUGNAX*¹

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Early in the present century, it was reported that a number of littoral organisms showed cycles of behavior which persisted under so-called constant laboratory conditions with tidal frequencies and with phases adaptively related to tidal events of the areas from which the organisms were collected (Gamble and Keeble, 1903, 1904; Bohn, 1904, 1906). Later, Gompel (1937) found that several species of animals display tidal rhythms of O_2 -consumption which also persist under constant laboratory conditions. These reports were not very successful in convincing the majority of biologists of the reality of persistent tidal rhythmicity. However, during the past few years, a number of studies have again pointed out that many organic processes, *e.g.*, color change, spontaneous activity, and O_2 -consumption, in a rather wide variety of plants and animals do indeed vary with primary lunar or tidal frequency under constant conditions.

Rao (1954) found that the filtering rate of species of *Mytilus* was greatest at the times of high tides in the areas of collection when the mussels were maintained in the laboratory. This rhythm of behavior was clearly apparent day by day. However, many of the lunar or tidal cycles that have been described are apparent only by statistical analyses of 15 or 29 days of continuous data (Brown, Freeland and Ralph, 1955; Brown, Webb, Bennett and Sandeen, 1955; Brown, Shriner and Ralph, 1956).

The results of the work to be described demonstrate that the fiddler crab, *Uca pugnax*, does have an overt rhythm of primary lunar frequency, a rhythm of spontaneous motor activity.

MATERIALS AND METHODS

In both 1955 and 1956, males of the species *Uca pugnax* were used in these studies. The crabs were collected from Champoquoit beach or Sippiwisset beach on the Buzzards Bay side of Cape Cod. Tidal events on these two beaches occur roughly 10 minutes later than they do at New York City. In the laboratory the animals were kept in white-enamelled pans in a small amount of sea water until they

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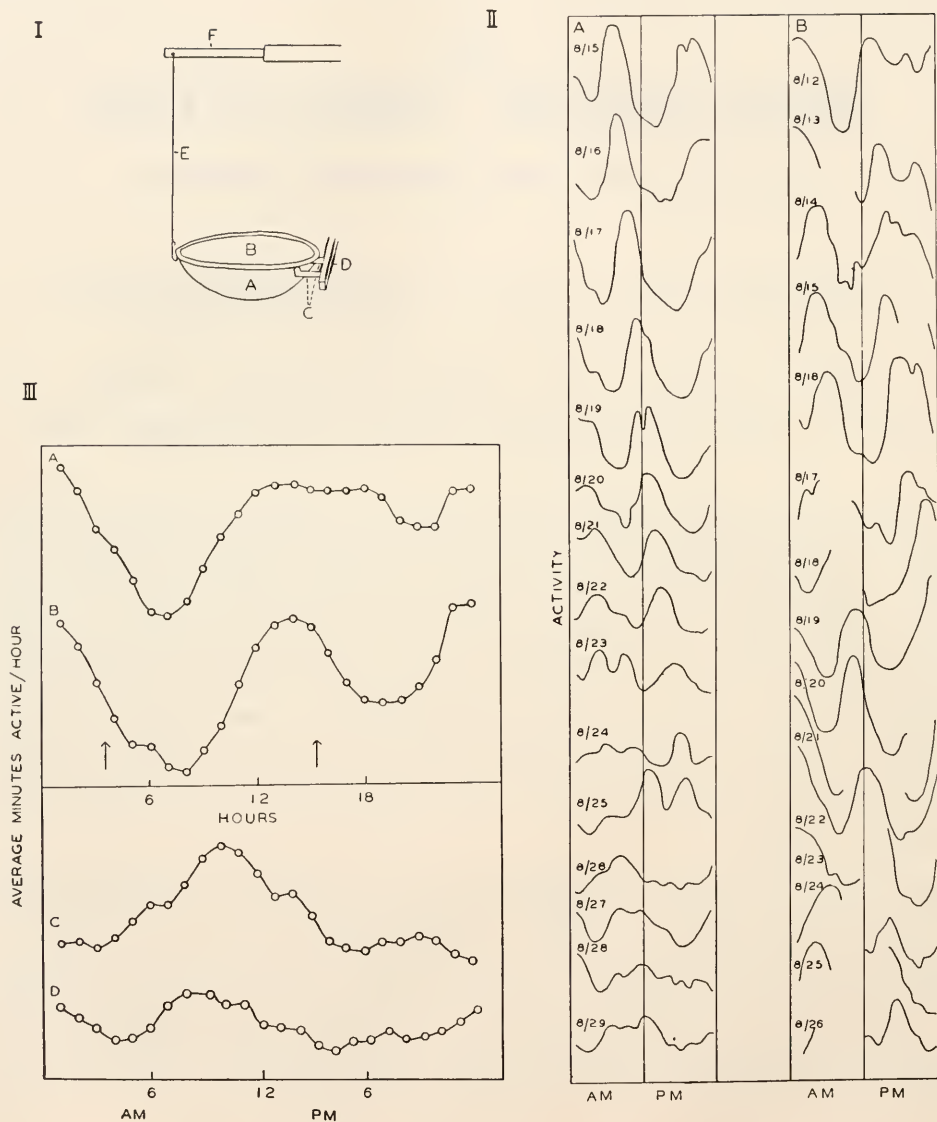


FIGURE 1. I. An illustration of part of the apparatus used to record the motor activity of an individual fiddler crab. For explanation of letters, see text. II. A, the average cycles of activity for a group of 20 crabs for the 15 consecutive days from August 15 through August 29, 1955. B, the average cycles of activity for a group of 20 crabs for the 15 consecutive days from August 12 through August 26, 1956. III. A and B, the mean, 29-day, tidal cycles of activity of fiddler crabs for July 6 through August 3, 1955 and August 2 through August 30, 1955, respectively. The arrows indicate the relative times of low tide at Chapoquoit Beach. C and D, the mean, 29-day, solar cycles of activity of fiddler crabs for July 6 through August 3, 1955 and August 2 through August 30, 1955, respectively.

were placed in the recording apparatus. This was done usually within two days of their collection.

Part of the recording apparatus employed is illustrated in Figure 1, I. A single crab was placed with a small amount of sea water in a plastic saucer (A), and covered with a circular piece of cardboard (B) which fitted the saucer tightly. Each of 10 saucers was supported on one side by metal bands (C) which were in turn fastened to a rigid horizontal bar (D). To the opposite side of each saucer was attached a nylon thread (E) which was fastened to the lever of a spring balance recording system (F) equipped with an ink-writing pen. The movements of the crabs in the finely balanced saucers were recorded on a kymograph which made one complete revolution every 24 hours. The spring balances and kymographs were of the type described by Brown (1954a).

The experiments were carried on in an inner room in a brick building at the Marine Biological Laboratory in which the light intensity at the level of the recording apparatus was at all times essentially constant and less than two ft. c. The containers with the inclosed crabs were shielded from movements and shadows in the laboratory. The air temperature in the room varied non-rhythmically from 21° to 24° C. through the summer months.

During the summer of 1955, the activity of 10 crabs was recorded continuously from July 6 through August 13, and that of 20 crabs was recorded from August 14 through 30. Freshly collected crabs were placed in the recorders on July 5, July 14, and August 14. In 1956, 20 crabs, which were collected on June 16 were placed in the recorders on that day, and their activity was recorded through the afternoon of June 26. On August 11, another group was collected, and the activity of 20 of these was recorded from that day through August 28.

The data recorded in 1955 were analyzed in the following manner: the number of minutes of each hour that each animal was active was determined from the kymograph recordings. From these figures was calculated the average number of minutes per hour that the population (either 10 or 20 individuals) was active. The hourly data were converted into three-hour moving means as given in Table I. Mean, 29-day solar and lunar cycles of activity were analyzed by methods used extensively in our laboratory (Brown, Bennett, Webb and Ralph, 1956; Brown, Freeland and Ralph, 1955) by which any possible solar or lunar cycles are synchronized day by day.

The results for 1956, given in Table II, are in terms of activity units per hour. Activity units were derived as follows: for each hour, the number of animals of the group of 10 that was active, *e.g.*, 8 out of 10, was recorded from the kymograph records. From these hourly values were calculated three-hour moving sums. The sums for the two groups of 10 each that were observed concurrently were added.

For the periods August 14–30, 1955, June 17–26, 1956, and August 12–28, 1956, when the motor activity of 20 individuals was recorded, the average values per hour for the two groups of 10 crabs each were correlated on an hour-by-hour basis. However, in this report, all data are given as the average for the entire population that was observed at any particular time.

RESULTS

The hourly values of spontaneous motor activity for *Uca pugnax* in 1955 are found in Table I, and those for the two periods of 1956 in Table II. By merely

TABLE II
The activity units/hour for groups of fiddler crabs for 1956

		Hour																								
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	
June	17	20	28	36	41	37	30	27	23	22	18	18	17	18	24	30	37	35	33	30	29	26	24	25	28	
	18	26	24	29	36	41	37	31	26	23	22	19	17	17	18	17	24	30	37	35	30	25	24	28	29	
	19	28	26	25	31	38	44	42	—	—	—	—	—	—	26	20	16	13	15	20	21	23	21	23	24	
	20	26	27	29	30	32	34	36	36	33	30	25	19	17	19	22	20	16	17	21	30	32	32	31	28	
	21	24	18	19	21	21	20	25	31	33	31	28	26	20	16	11	12	10	13	19	26	29	30	28	28	
	22	27	26	25	22	24	23	24	28	31	33	32	33	28	20	14	17	23	23	22	24	30	30	26	25	
	23	27	29	27	26	29	28	25	24	30	42	42	40	30	25	20	18	16	12	9	13	22	23	30	27	
	24	26	25	24	21	24	24	24	21	24	32	33	31	28	23	18	11	10	9	10	8	19	26	35	30	
	25	28	25	27	27	27	24	24	21	21	25	29	30	28	25	17	12	7	11	14	12	11	14	26	34	
	26	33	30	29	27	27	26	23	19	15	20	23	27	27	26	20	16	11	—	—	—	—	—	—	—	
	Aug.	12	57	56	53	51	43	35	28	27	25	28	37	50	55	56	54	50	48	47	52	50	48	44	49	54
		13	58	56	53	48	44	—	—	—	—	36	32	39	48	52	49	42	41	40	43	46	42	35	29	29
14		32	38	48	49	49	40	35	24	24	21	30	28	33	37	42	47	44	44	39	40	40	37	31	24	
15		28	37	46	—	—	—	36	29	28	24	18	18	22	32	41	47	45	38	—	—	—	—	38	32	
16		24	20	24	32	37	40	35	27	16	13	13	11	9	9	15	29	42	44	43	39	43	36	26	20	
17		17	26	24	29	29	—	—	—	—	22	17	13	13	14	10	7	14	24	30	31	27	26	22	21	
18		15	13	13	17	21	25	—	—	—	—	—	13	9	11	11	13	12	14	18	27	35	42	40	34	
19		24	20	16	14	8	8	10	20	28	30	31	26	20	14	11	10	10	12	17	20	25	34	41	40	
20		34	27	22	16	14	14	16	25	34	40	36	29	20	14	8	7	6	12	14	—	—	—	44	41	
21		39	35	29	24	17	13	9	10	—	—	—	—	—	—	—	—	14	9	7	7	11	23	35	35	
22		38	34	26	23	19	16	10	10	15	23	28	32	31	29	23	16	11	8	10	10	11	16	23	30	
23		36	35	34	27	24	20	21	17	17	17	19	—	—	—	39	21	15	13	12	10	11	13	28	28	
24		19	24	28	32	36	36	36	31	—	—	—	15	14	20	22	26	20	15	11	11	9	12	12	17	
25		21	28	33	33	29	25	—	—	—	—	13	12	—	—	32	30	22	18	13	13	11	11	11	11	
26		16	20	23	—	—	—	—	—	—	—	—	22	22	19	23	29	34	33	28	23	21	17	17	19	
27		25	25	27	26	31	32	38	37	32	30	28	25	19	16	22	23	25	25	27	27	26	23	20	17	
28		16	18	21	20	25	27	32	32	33	29	—	—	—	—	—	—	—	—	—	—	—	—	—	—	

minutes per hour active in 1955, and 8 activity units per hour to 58 activity units per hour in 1956. Near the end of a particular recording period, both the mean daily activity and the amplitude of the cycles are lower than at the beginning of the same period.

These results are shown graphically in Figure 1, II A and B, in which are plotted the cycles of motor activity for groups of 20 crabs for August 15 through 29, 1955 and August 12 through 26, 1956. In Figure 1, II A, one can follow the moving of the two peaks of activity across succeeding solar days from August 15 to 22. In this instance, the peak that occurred between hours 7 and 8 on August 15 occurs between hours 14 and 15 on the 22nd, and the evening peak of August 15 has moved to the early morning on August 22. For the cycles of August 23 through 29, it is more difficult to distinguish maxima or the movement of these maxima very precisely. There seems to be a warping and diminution of a peak as it moves into the afternoon and evening hours. The peak of the morning hours which can be identified on August 20, can be traced through August 29; however, it, too, shows warping and broadening.

Generally, the same characteristics are to be seen in Figure 1, II B, especially for the cycles of August 12 through 16. After this time, the data are incomplete, and it is not advisable to complete the curves from only the available data. A period of high activity seen just after 0 hour on August 12 is identified as a rather sharp maximum at 12 on August 22, and similarly the afternoon high (hour 14) on August 12 is identified between hours 3 and 4 on August 25. Again, the warping and broadening of maxima, observed in 1955, are evident.

In order to demonstrate the reality of the movement of peaks of activity across solar days at the average primary lunar or tidal rate of 51 minutes per day, the rate of movement for specific peaks was determined for 20 different intervals from the data of 1955 and 1956. The intervals used were in no case less than two days or more than 11 days. The average rate for these periods was 50.1 ± 3.58 minutes per day.

Tidal phase relationships as well as frequency relationships are to be noted in these rhythms of activity. It was observed that in 1955, the peaks of activity occurred two to three hours before the times of low tide on the beaches from which the crabs were collected. In 1956, the maximum activity was, on the average, four to five hours before low tides.

This phase relationship is shown in Figure 1, III in which are plotted the mean, 29-day tidal cycles of locomotor activity for July 6 through August 3, 1955 (A) and August 2 through 30, 1955 (B). These curves are plotted so that the times of low tides at Chapoquoit Beach, as indicated by arrows, lie directly under one another, although the tidal events actually occurred at different times on the two days with which the 29-day analyses were begun. The times of low tide were: 3:22 and 15:19 on July 6 and 1:34 and 13:37 on August 2. In these figures, the form of the tidal cycle, discussed previously, is again apparent. In A, a sharp peak of activity is seen at hour 1, or 2 hours and 22 minutes before low tide. The second maximum takes the form of a broad peak from hour 13 through 18, extending from 2 hours before until 3 hours after low tide. Activity fell rather steeply from 1 until 7 while the activity following the second maximum did not decrease so steeply nor to so great a degree. The cycle for August 1955 (Fig. 1, III B) was very much like that illustrated in Figure 1, III A, although the second period of high activity does not last so long in the former as in the latter. Again, the activity persisted at a higher level following the second maximum than it did following the first peak.

In Figure 1, III C and D are plotted the mean, 29-day, solar cycles for the same periods of time as the lunar cycles. It is apparent that the amplitude of the solar cycles is less than that of the lunar cycles. From the lowest to the highest values there was a 2.4-fold increase in the tidal cycle for July 6 through August 3, and a 2.2-fold increase in the tidal cycle for August 2 through 30, whereas the increases for the solar cycles for the same periods were 1.6-fold and 1.3-fold, respectively. Although the amplitude of these solar cycles is low, the cycles indicate that activity between hours 6 and 12, other factors equal, is higher than at other times of the solar day. This tendency can be seen in Figure 1, II A by comparing the heights of the two peaks, *i.e.*, the morning peak is higher than the afternoon peak from August 15 through 17.

Coefficients of correlation for the activity of two groups of crabs, the activities of which were recorded independently during the same periods of time, were: $+0.768 \pm 0.028$ for August 14 through 30, 1955; $+0.410 \pm 0.058$ for June 17 through 26, 1956; and $+0.687 \pm 0.034$ for August 12 through 28, 1956.

DISCUSSION

The cycle of spontaneous motor activity for the fiddler crab, *Uca pugnax*, described in this report, appears to be the first example of a clearly overt locomotor rhythm of primary lunar or tidal frequency. The occurrence of two peaks of ac-

tivity, 12 to 13 hours apart, within one solar day, and the movement of these peaks across succeeding solar days at an average tidal rate establish the reality of a tidal cycle persisting under laboratory conditions. It must be pointed out that the day-to-day preciseness of this rhythm decreases somewhat after the crabs have been in the recording containers seven or eight days. The warping and broadening of peaks, as well as the diminution of activity, discussed previously, cannot be explained satisfactorily at the present time. It is tempting to postulate that these phenomena are indications that an internal timing mechanism is not able to maintain a precise cycle longer than a week or so under constant conditions, and that after this time unknown external signals alone maintain only a less regular rhythm. Evidences for both endogenous and exogenous components of persistent rhythmicity in fiddler crabs have been reported recently (Brown, Webb and Bennett, 1955; Brown, Webb, Bennett and Sandeen, 1955).

The difference between the phase relationships of actual tidal events and the peaks of activity observed in 1955 and 1956 brings up questions regarding the setting of phases of tidal cycles, and to what extent behavior cycles under constant conditions parallel those of the organisms in their natural environments. The lunar or tidal cycles described previously indicate that although many species have cycles of 12.4 and/or 24.8 hours, phase relationships are species specific. For example, the spontaneous activity of quahogs is low during the times of low tides when this species may not be covered by water (Bennett, 1954), while the pigment in the melanophores of the fiddler crab, *Uca pugnax*, is most dispersed shortly before the times of low tide when these animals are typically active on the beaches (Brown, Fingerman, Sandeen and Webb, 1953). In these cases, the phases of the persistent tidal cycles seem to indicate some adaptiveness of the cycles to conditions which obtain under natural field conditions. On the other hand, Fingerman (1956) has reported that fiddler crabs, *Uca pugilator* and *Uca speciosa*, collected from regions of the Gulf coast where there is but one low tide per day, have persistent tidal cycles of color change characterized by two peaks of pigment dispersion during each solar day, one shortly after low tide, and the second 12.4 hours later or shortly after high tide.

The relationship observed between the fiddler crab activity and the times of low tide in 1955, *i.e.*, maximum activity two to three hours before low tides, might suggest that the behavior of the crabs in the laboratory is much the same as that of the crabs on the beaches. Typically, these crabs begin to emerge from their burrows as the water recedes following a high tide, and great numbers of these crabs are usually running on the beaches until shortly before low tide. However, the observations for 1956, *i.e.*, that the peaks of activity occurred four to five hours before low tide, suggest that the phase relationships of persistent tidal cycles change, and may not always reflect adaptive behavior of the species in its natural, changing, physical environment. It is possible that the difference in the rhythmic behavior of the crabs between the two years may reflect differences noted in field observations between these same two years. In 1955, as was usual, the crabs were collected easily shortly before the times of low tides. In 1956, the animals were difficult to collect since there were few running on the beaches before low tides. Many of the crabs used in 1956 had to be dug from their burrows. It is possible that stimuli other than those resulting from emergence and running of the crabs may set the phases of the observed tidal cycles.

That the phases of a tidal cycle do shift in the absence of experimental modifications is shown clearly in the report of Brown (1954b). In this work, it was found that the maxima of the tidal cycle of oysters collected from New Haven Harbor and shipped to Evanston, Illinois correlated with the times of high tide in the native habitat during the first two weeks of maintenance under constant conditions. During the next month, the same group of oysters showed a rhythm with maxima at the times of lunar zenith and nadir. Two other reports contain information regarding the experimental shifting of the phases of tidal cycles (Brown, Fingerman, Sandeen and Webb, 1953; Rao, 1954). However, since the problem of the setting of phases of persistent tidal cycles is one that has not been investigated to a great degree as yet, much more work must be done before any definite statements can be made.

The overt tidal rhythm described here promises to be one by which not only the problem of phase setting but also experimental modifications of tidal cycles can be studied. The facts 1) that the cycle of locomotor activity repeats itself rather precisely on a day-to-day basis for at least a week after the crabs are placed under constant conditions, and 2) that the cycles of two independent small groups correlate significantly to a high degree do point to its usefulness in such studies.

SUMMARY AND CONCLUSIONS

1. The spontaneous locomotor activity of groups of fiddler crabs, *Uca pugnax*, was recorded during the summers of 1955 and 1956 under constant laboratory conditions.

2. This species shows an overt rhythm of activity of primary lunar or tidal frequency. Within solar days, there are two peaks of activity which are 12 to 13 hours apart. These maxima move across succeeding solar days at an average tidal rate. The cycles are precise for at least a week under constant conditions, but after this time, some warping and displacement of maxima occur.

3. A low amplitude solar rhythm of activity is apparent upon analysis of 29 days of continuous data. This rhythm is characterized by high activity between hours 6 and 12 of the solar day.

4. There was observed a difference in phase relationships of the tidal rhythms between 1955 and 1956. The state of the problem of the setting of phases of persistent tidal cycles is discussed.

5. Since this rhythm is precise for at least a week under constant conditions, and since the cycles of two groups of crabs recorded independently correlate to a high degree, this cycle appears to be an excellent one with which to study experimental modifications of persistent tidal rhythms.

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